Remarks

Claims

Claims 1-40 were pending in the prior application. These claims were rejected under 35 U.S.C. § 112 first paragraph and under 35 U.S.C. § 103 for the reasons set forth in the Office Action mailed on March 21, 2001 (Paper No. 12) (herein referred to as "Office Action"). Applicant respectfully requests that the Examiner consider the pending claims in light of the following discussion of prior claim rejections under 35 U.S.C. § 112, first paragraph.

Rejections Under 35 U.S.C. § 112 Second Paragraph

The Examiner rejected claims 1-17 and 23-25 under 35 U.S.C. § 112, second paragraph. Applicant has made a number of clarifying amendments which retain a focus on the subject matter of the invention and address a number of the Examiner's concerns as well. At the same time, however, some aspects of the rejection are not well taken. Accordingly, this ground of rejection are respectfully traversed.

Claims 1 and dependent claims - "...at said locations of the human skin wherein the chimeric RNA-DNA oligonucleotide has a double hairpin structure with pyrimidine loops"

Claims 1and 18, and claims depending therefrom stood rejected as indefinite because of the recitation of the clause "wherein the chimeric RNA-DNA oligonucleotide has a double hairpin structure with pyrimidine loops" following the language "...at said locations of the human skin". The Examiner asserts that it "unclear whether the resultant phenotypic changes only occur in location where the RNA-DNA oligonucleotide has a double hairpin structure with pyrimidine loops or whether the changes occur in locations whereto the RNA-DNA oligonucleotide has been delivered." Applicant disagrees.

Applicant respectfully submits that there are two "wherein" clauses in claims 1 and 18. The first "wherein" clause further defines the composition and the second "wherein" clause further defines the chimeric RNA-DNA oligonucleotide already recited in the first clause. There is nothing indefinite about further defining claim elements using "wherein" clauses. However, without conceding the validity of this rejection and solely to facilitate prosecution, claims 1 and

18 have been amended by adopting the language suggested by the Examiner. Accordingly, it is respectfully submitted that the present claims 1 and 18, and the dependant claims are not indefinite and requests withdrawal of this rejection.

Claims 8-10 and 23-25 "contiguous nucleotides in each of the first and second strings"

Claims 8-10 and 23-25 stood rejected as indefinite because of the phrase "contiguous nucleotides in each of the first and second strings". The Examiner averred that it is not clear whether the "contiguous is directed to the structural relationship existing between the first and second strings or whether it merely recites the fact that each of the two strings contain a set of contiguous nucleotides, which may or may not be contiguous with those of the other string." Applicant respectfully submits that the rejected claims, as amended, clarifies that each of the two strings contain a set of contiguous nucleotides, thereby obviating this ground of rejection.

Claims 8-10 and 23-25 - "nuclease protected"

Claims 8-10 and 23-25 stood rejected based on the assertion that these claims are indefinite in their recitation of "nuclease protected". Applicant respectfully submits that the relevant claims have been amended to more clearly claim the subject matter of the invention. Further, it is respectfully submitted that the guidance in the specification, for example, at page 32, line 10 through page 34, line 20 provides sufficient description of what nucleases the oligonucleotide is resistant to. For example, at page 32, line 21, the specification teaches that the oligonucleotide is resistant to the RNaseH and other RNases. See, also p. 34, line 15. The nucleotides in loops are also made resistant to RNases as long as these nucleotides are ribose nucleotides. Notwithstanding, solely to facilitate prosecution, Applicant amend claims to specify that these are RNase protected. Reconsideration and withdrawal of this rejection are respectfully requested.

As to whether the first and second strings are contiguous, Applicant respectfully submits that one skilled in the art would know from the guidance in the specification and from a reading of independent claim 1 or 18 which set forth the metes and bounds of chimeric RDOs. For example, there are a number of examples of chimeric RDO sequences have been described throughout the specification. All of them show a double-hairpin structure containing pyrimidine loops (made of either "T" residues or "U" residues), and 3' and 5' unligated ends (see also

specification at p. 32, line 29). Claim 1 or 18 clarifies that the chimeric RNA-DNA has a double hairpin structure with pyrimidine loops. Claims 8-10 or 23-25 clarify that the first and second strings are fully or substantially complementary to each other. Double hairpin structure of the chimeric RDO is possible because of these complimentary or substantially complementary first and second strings, and the pyrimidine loops. Applicant respectfully submits that these teachings and claim recitations are sufficient to reasonably apprise those of ordinary skill in the art as to whether or not the first and second strings are contiguous or have some intervening sequences between them.

Accordingly, Applicant respectfully submits that withdrawal of the rejections under 35 U.S.C. §112, second paragraph is in order.

Rejections Under 35 U.S.C. §112, First Paragraph

Claims 2-4, 19, 32 and 33 stood rejected under 35 U.S.C. § 112, first paragraph for lacking sufficient written description in the specification *i.e.*, as containing subject matter which was not described in the specification in such a way as to reasonably convey to one skilled in the relevant art that the inventor(s), at the time the specification was filed, had possession of the claimed invention. Applicant respectfully traverses for the reasons set forth below.

The Examiner contended, among other things, that the specification does not provide an adequate written description of sequences of genes embraced by claims 4, 19, and 33. Claim 4, 19 or 33 recites a total of 60 genes. Applicant, in its response filed on December 20, 2000, has provided evidence of sequences for a total 8 genes recited in these claims; seven gene sequences available from the GenBank (GenBank accession #4502960 for COL7A1, #6678659 for LAMA3, #4557712 for LAMB3, #9845499 for LAMC2, #4557674 for ITGA6, #4504768 for ITGB4, #4505876 for PLEC1) and one gene sequence (KRT14) disclosed in Coulombe et al., 1991, Cell, 66:1301-1311, which reference is of record. The Examiner has correctly noted that one of these eight genes is from the mouse (LAMA3). The other seven are human genes. Thus, the Applicant has provided the sequence information for seven human genes. Applicant submits herewith a table (as Exhibit 1) showing the GenBank numbers for human sequences for all of the genes recited in claims 4, 19, 33. The GenBank information is public information. One skilled

in the art would understand how to design chimeric RDOs based on the publicly available sequence information in order to carry out the claimed invention. In other words, when the present specification is read in light of the knowledge and level of skill in the art, the specification describes the claimed methods. Therefore, the disclosure obligation under the "description requirement" of § 112, first paragraph, is satisfied. Applicant is not required to describe every detail of the invention. *In re Hayes*, 25 USPQ2d 1421 (Fed. Cir. 1992).

At page 4 of the Office Action, the Examiner asserts that the references cited by the Applicant in Table 1 on pages 51 and 52 of the application do not report the sequences that would be used to design chimeric RDOs. The Examiner offers no specific support for this assertion, and offers no examples of different references. In fact, Table 1 includes citations to articles which truly do report such sequences sufficient for designing chimeric RDOS. (See, for example, Ahmad et al., 1998, Science, 279:7201.; Armstrong et al., 1999, Hum Molec Genet 8:143; Dong et al., 1998, Cancer Res. 58:3787; Frank et al., 1999, Nature 398:473; Korge et al., 1998, J Invest Dermatol 111:896; Marsh et al., 1997, Nat Genet 16:333; McGrath et al., 1997, Nat Genet 17:240; Richard et al., 1998, Hum Genet 103:393; Richard et al., 1998 Nat Genet 20:366; Rickman et al., 1999, Hum Mol Genet, (In Press); Rowan et al., 1999, J Invest Dermatol 112:509; Sakuntabhai et al., 1999, Nat Genet 21:271; Winter et al., 1997, Nat Genet 16:372. Copies of these references are provided as part of the Information Disclosure Statement and Form PTO-1449 which are being submitted concurrently herewith. It should be noted that some of these references also provide GenBank Accession Numbers for the sequences. It is hoped that, upon inspection of these references, the Examiner will withdraw all rejections based upon his erroneous assumption concerning Table 1. If the Examiner seeks to justify based on his own knowledge, that the prior art made available herein does not provide sequence information sufficient for designing chimeric RDOs, the Examiner must submit a declaration as required by 37 CFR § 104(d)(2).

The Examiner further rejected claims 1-40 under 35 U.S.C. § 112, first paragraph. The Examiner asserts that the specification does not enable one skilled in the art to make and use the

invention commensurate in scope with these claims. Applicant respectfully traverses this rejection.

The Examiner continues to rely on Stephenson (JAMA 281(2):119-120, 1999) and Strauss (Nature Medicine, 4(3): 274-275, 1998) to assert that the invention is drawn to a highly unpredictable art. Applicant respectfully submits that this assertion is unwarranted because, as already pointed out by the Applicant in its last Office Action response filed on Dec 20, 2000, these references do not comment on the others' success or failures relating to the modification of genes of a skin *in vivo* using chimeric RDOs. Notwithstanding this fundamental difference, the Examiner asserts on page 7of the Office Action, that "Applicants have failed to provide a sufficient explanation as to why Applicants approach has not been shown to reproducibly work in other laboratories." The Examiner acknowledges the publication of Kren (Kren et al., 1998, Nature Med., 4:285-290) that reports the successful use of this technique. Appropriately understanding that Kren's work undercuts the reasoning behind the instant rejection, the Examiner cites a commentary by Strauss (Nature Medicine, 4: 274-275, 1998) that speculates about the merits of Kren's work.

As a preliminary matter, one must note that the Kren publication is a report of Kren's actual work, while Strauss merely speculates, without even attempting to reproduce Kren's work. Kren's successful work passed the muster of peer-review before its publication. Thus, as against Kren, Strauss' comments on Kren's paper are entitled to little weight.

However, the validity of Kren's actual results in the face of Strauss' speculations are supported by more than Strauss' own lack of basis for his statements. Kren's group has successfully demonstrated the correction of the UDP-glucuronosyltransferase gene defect using chimeric RDOs (Kren et al., 1999, Proc. Natl. Acad. Sci. 96:10349-10354, a copy of this reference accompanies this amendment). This work was published after the publication of Strauss' comments about Kren's prior work. Further, Kren's work has been expanded and favorably commented upon by others who actually work on this technology. See, for example, Bandyopadhyay et al., 1999, Journal of Biological Chemistry, 27:10163-10172 and Bartlett et al., 2000, Nature Biotechnology, 18:615-622, copies of which are enclosed herewith as Exhibit 2. Thus, as of well before the filing date of this application, Kren proved the usefulness of this

technique. The enabling nature of Kren's work has been confirmed by the actual laboratory work of others in this art. Strauss' mere speculation is thus shown to be uninformed and in error. No rejection based on Strauss' speculations contradicting Kren can stand.

Further, Applicant's claims relate to *in vivo* gene modifications in skin tissue. Because the skin tissue is different from tissues such as liver and because the references cited by the Examiner do not relate to *in vivo* gene modifications in skin tissue, the Examiner has not met the burden of establishing that the Applicant's invention is not enabled. The skin tissue is different from tissues such as liver in that the target cells in skin tissue comprise epidermal stem cells. It is well known in the art that the epidermal stem cells are undifferentiated, pluripotent, and slow-cycling cells with an ability to proliferate throughout the life span. (See, Latkowski et al., 1999, Epidermal cell kinetics, epidermal differentiation, and keratinization, Chapter 9, in: Fitzpatrick's Dermatology in General Medicine, Freedberg et al., eds, McGraw-Hill; Lavker et al., 1999, Biology of hair follicles, Chapter 17, in: Fitzpatrick's Dermatology in General Medicine, Freedberg et al., eds, McGraw-Hill. Copies of these references are enclosed herewith as Exhibit 3. The stem cells have the capacity for self-renewal and the ability to generate progeny of cells called transit amplifying cells. The transit amplifying cells are rapidly dividing cells which go through a limited number of cell divisions before entering the differentiation pathway. Thus, in skin tissue, each stem cell divides and gives rise to a daughter stem cell and a transit amplifying cell. The transit amplifying cells become the terminally differentiated cells. These scientific facts are well known to one skilled in the art. The gene modification even in few stem cells would result in an apparent high level of genetic modifications and phenotypic changes as these stem cells continue to expand and repopulate the epidermis. Neither Strauss nor Stephenson comment upon *in vivo* gene modifications in skin tissue. Therefore, the Examiner's contentions based on these articles fail and thus the Examiner has not established why the specification does not realistically enable one skilled in the art to practice the invention to the scope claimed.

As to Applicant's statements in Stephenson article (at p.120, left column, last paragraph), it has already been pointed out that these statements relate to the ability of RDO's to modify genes in cells *in vitro*, not *in vivo* comprising the target cells described in the above paragraph. The Examiner now asserted on page 7 of the Office Action that because Applicant "has not

adequately resolved the question of why [its] method works so well in vivo in contrast to *in vitro*, it underscores the lack of predictability in this undeveloped art." The Examiner conceded that Applicant method works "well in vivo". The claims are limited to *in vivo* methods not *in vitro* methods. The enablement requirement demands that the patent specification enable one skilled in the art to make and use only the claimed invention without undue experimentation. *Genentech, Inc., v. Novo Nordisk*, 42 USPQ2d 1001 (Fed. Cir. 1997). Applicant has done so. It is not a requirement of §112 that the Applicant provide an explanation, or even know, how or why the invention works so well. *Newman v. Quigg*, 11 USPQ2d 1340, 1345 (Fed. Cir. 1989). Under the law, the Applicant need not comprehend the scientific principles on which the practical effectiveness of its invention rests. In fact, the Federal Circuit recently reaffirmed that the enablement prong is met if one satisfies, as Applicant does here, the requirement of teaching how to achieve the claimed result, even if the theory of operation is not correctly explained or even understood. *In re Cortight*, 49 USPQ2d 1464 (Fed. Cir. 1999).

The Examiner's unsupported reasoning that "cells cultured *in vitro* would be expected to more closely resemble, at a metabolic level, the ... epidermal stem cells in contrast to the predominant cell type in skin that is quiescent, non-dividing, and postmitotic" is not well taken. Cells cultured *in vitro* by the Applicant were melanocytes, not epidermal stem cells. See Alexeev et al., 1998, Nature Biotechnology, 16:1343-1346. The melanocytes cannot be induced to proliferate *in vitro* to the same extent as epidermal stem cells. Further, it is well known that the cells *in vitro* cannot be provided with the same growth conditions available to the cells *in vivo* under the current state of the art. If the Examiner is aware of documents supporting that the melanocytes cultured *in vitro* resemble epidermal stem cells *in vivo* as to their proliferation rates, the Examiner is requested to cite them. If not, he is requested to provide his own Declaration under 37 CFR §1.104(d)(2) or withdraw the unsupported assertion.

Applicant has shown that gene modification in skin cells *in vivo* can be achieved by practicing the claimed invention and, as conceded by the Examiner, its method "works ... well in vivo". Therefore, Applicant respectfully maintains that the lack of similar degree of success with gene modification in melanocytes *in vitro* or tissues such as liver does not support a conclusion

that the claimed invention which relates to the genetic modification in skin *in vivo* is nonenabled. Alleged lack of enablement of unclaimed subject matter is not a basis for §112 rejection.

The Examiner also averred that the disclosure is not enabled for genetic modifications involving insertions or deletions. Applicant respectfully disagrees and submits that it has provided working examples for making genetic modifications using chimeric RDOs to cause substitutions in skin cells. The genetic modifications using chimeric RDOs to cause insertions or deletions in skin cells are further embodiments. These further embodiments can be made by simply following the teachings in the specification and the general knowledge available to one skilled in the art. For example, the specification teaches how a deletion can be made in a DSG3 gene. See, page 18, line 18 through page 19, line 29. The Examiner suggests that while claims to gene modifications involving substitutions by RDO are enabled *in vivo* but not those involving insertions or deletions. The Examiner has not established why gene modifications involving insertions or deletions are not enabled. Applicant respectfully submits that oligonucleotide mismatch mutagenesis is a popular method known to one skilled in the art and is used to create a desired mutation at a predetermined site within a DNA molecule. Skilled artisans using this RDO method have already shown that it is possible to delete or insert nucleotides that will result in deletion or insertion of amino acids (See, Kren et al. 1999, PNAS 96:10349-10354; Cole-Strauss, A. et al. 1999 Nucleic Acids Res. 27:1323-1330. Copies of these references accompany this amendment as Exhibit 4). Based on this knowledge and the guidance in the specification, one skilled in the art could have genetically modified skin cells using chimeric RDOs to cause insertions and deletions in skin cells simply by carrying out routine experimentation. To satisfy the enablement requirement, Applicant need not describe all actual embodiments *SRI Int'l v. Matsuhita*, 775 F.2d 1107 (Fed. Cir. 1985). Therefore, Applicant respectfully submits that the specification is replete with teachings enabling a person skilled in the art to which the invention pertains, or with which it is most nearly connected, to make or use the invention commensurate with the scope of these claims.

The Examiner further asserts that the evidence of record does not provide sufficient nexus for concluding that the claimed methods, as recited in the mouse models, can be readily extrapolated to humans and cites Crystal (1995), Science 270:404-410 at p. 409 bottom, left

column. More specifically, the Examiner points to the following language: "[H]umans are not simply large mice. There have been several surprise examples, in which predictions from gene transfer studies in experimental animals have not been borne out in human safety and efficacy trials." The language in Crystal pointed out by the Examiner relates to human safety and efficacy trials. Applicant respectfully submits that this requirement, i.e., reduction of invention to practice to the extent of human safety and efficacy is contrary the present case law. *In re Brana* 51 F.3d 1560, 1567-1568 (Fed. Cir. 1995). Applicant respectfully submits that its invention has been enabled in an art-recognized animal model. (For art recognized status of mice, See Larker et al., 1999, Biology of hair follicles, Chapter 17, in: Fitzpatrick's Dermatology in General Medicine. A copy of this article is made part of Exhibit 3. See also, Heinonen et al., 1999, J. Cell Science 112:3641-3648; Vassar et al., 1997, Cell 64:365-380. Copies of these references accompany this amendment as Exhibit 5). Under the law, one may establish the feasibility of a particular application in question in a "standard experimental animal" that is routinely used as a model system by those skilled in the art, without any requirement for clinical testing on humans; one skilled in the art is expected to make further correlation by him or herself. *In re Krimmel* 292 F.2d 948, 953-954 (CCPA 1961); Indeed, testing for full efficacy in humans, now that the inventive principle has been demonstrated by the Applicant, is appropriately left for the Food and Drug Administration. *Scott v. Finney*, 34 F.3d 1058 (Fed Cir. 1994). The Title 35 does not demand that such human testing occur within the confines of Patent and Trademark Office (PTO) proceedings. Therefore, the Examiner's rejection of the claims is improper.

The Examiner also asserted that there is insufficient guidance concerning the threshold levels of correction required to generate a phenotypic change or to constitute a measurable phenotypic change. Applicant respectfully submits that one practicing the invention would look for a phenotypic change in a treated animal skin by comparing it to the controls. For example, figure 3 shows albino mice, both treated and untreated. The hairs of Tyr-A RDO (test) treated mice were compared with the hairs of Tyr-B RDO (control) treated mice. The phenotypic change in the Tyr-A RDO is a result of gene correction. Further, for example, a composition comprising about 10 μg of RDO when injected intradermally was sufficient to bring about genetic changes in three different genes in the skin cells (see Examples 1 and 4) and to cause

phenotypic changes. Importantly, skin disorders such as the ones listed in Table 1 allow the gene correction to be identified phenotypically, a feature that could enable a valid measure of frequency. From this guidance and by carrying out routine experimentation, one skilled in the art can determine gene correction by simply looking at the phenotypic changes during the treatment of other genetic disorders. Thus, the specification provides reasonable guidance to enable those skilled in the art to make and use the full scope of the claimed invention.

Based on the above arguments, Applicant respectfully submits that the specification sufficiently discloses the invention, as presently claimed, so that one skilled in the art can practice the invention as claimed without resorting to undue experimentation. Reconsideration is respectfully requested.

In view of the foregoing and the amendments presented herein, Applicant respectfully requests that the Examiner reconsider and withdraw the rejection of claims under 35 U.S.C. §112, first paragraph.

Conclusion

A favorable reconsideration in view of the above remarks and allowance of the pending claims in the application are earnestly solicited.

Respectfully submitted,

Date: September 21, 2001

William J. McNichol, Jr.
Registration No. 31,179
Attorney for Applicant

REED SMITH LLP
2500 One Liberty Place
1650 Market Street
Philadelphia, Pennsylvania 19103-7301
Fax: (215) 241-7945
Attn: William J. McNichol, Jr., Esq.
(215 241-7950)
Nanda P.B.A. Kumar, Esq.
(215 241-7991)

Marked Up Version of Claims in Serial No. 09/473,872 in response to the Office Action of March 21, 2001

1. (Twice Amended) A method of modifying a selected gene in cells of a human skin [at one or more locations] in vivo which comprises delivering to said cells at one or more locations of the human skin an effective amount of a composition sufficient to bring about stable genetic and phenotypic modifications in the selected gene at said locations wherein the composition comprises a chimeric RNA-DNA oligonucleotide having a double hairpin structure with pyrimidine loops and a pharmaceutically acceptable carrier such that said genetic modifications are made to the selected gene which result in phenotypic changes at said locations of the human skin [wherein the chimeric RNA-DNA oligonucleotide has a double hairpin structure with pyrimidine loops].

2. (Amended) The method of claim 1, wherein the stable genetic modification is in an epidermal fragility disorder gene selected from the group consisting of COL7A1, LAMA3, LAMB3, LAMC2, COL17A1, ITGA6, ITGB4, PLEC1, KRT5, KRT14 and PKP1.

3. (Amended) The method of claim 1, wherein the stable genetic modification is in a keratinization disorder gene selected from the group consisting of KRT1, KRT10, KRT9, KRT16, LOR, KRT2e, KRT6a, KRT 16, KRT 17, STS, TGM1, GJB2, GJB3, ATP2A2, DSP and DSG1.

4. (Amended) The method of claim 1, wherein the selected gene is tyrosinase, COL7A1, LAMA3, LAMB3, LAMC2, COL17A1, ITGA6, ITGB4, PLEC1, KRT5, KRT14, PKP1, KRT1, KRT10, KRT9, KRT16, LOR, KRT2e, KRT6a, KRT 16, KRT 17, STS, TGM1, GJB2, GJB3, ATP2A2, DSP, DSG1, HR, hHB1, hHB6, PAX3, TYR, TYRP-1, OCA2, OA1, MITF, HPS, FECH, UROS, URO-D, XPA, XPB, XPC, XPD, XPG, [CSB,] PTC, STK11/LKB1, PTEN, PTEN, XPB, XPD, WHN, GLA, ATM, ENG, ALK-1, PPO, BPAG2, or DSG3 gene.

8. (Twice Amended) The method of claim 1, wherein the chimeric RNA-DNA oligonucleotide comprises:

(a) a first string of nucleotides wherein the first string is made of at least four contiguous deoxyribonucleotides flanked on each side by at least nine ribonucleotides; and

(b) a second string of deoxyribonucleotides that is fully complementary to the first string of nucleotides, and

wherein the chimeric RNA-DNA oligonucleotide is, [nuclease] RNase protected, and wherein the chimeric RNA-DNA oligonucleotide has [contiguous nucleotides] in each of the first and second strings a set of contiguous nucleotides that are fully complementary to a segment of DNA of the selected gene except that the first string has one mismatching deoxyribonucleotide in said contiguous deoxyribonucleotides that defines a site of modification in the selected gene.

9. (Twice Amended) The method of claim 1, wherein the chimeric RNA-DNA oligonucleotide comprises:

(a) a first string of nucleotides wherein the first string is made of at least 20 ribonucleotides; and

(b) a second string of deoxyribonucleotides having the same number of deoxyribonucleotides as in the first string of nucleotides, wherein the second string is fully complementary to the first string of nucleotides except that the second string has a deoxyribonucleotide that forms a mismatched base pair with the corresponding nucleotide in the first string, and

wherein the chimeric RNA-DNA oligonucleotide is [nuclease] RNase protected, and wherein the chimeric RNA-DNA oligonucleotide has [contiguous nucleotides] in each of the first and second strings a set of contiguous nucleotides that are fully complementary to a segment of DNA of the selected gene except that the deoxyribonucleotide in the second string also forms a mismatched base pair with the corresponding deoxyribonucleotide in the DNA strand of the selected gene which mismatched base pair defines a site of modification in the selected gene.

10. (Twice Amended) The method of claim 1, wherein the chimeric RNA-DNA oligonucleotide comprises:

(a) a first string of nucleotides wherein the first string is made of at least four contiguous deoxyribonucleotides flanked on each side by at least nine ribonucleotides; and

(b) a second string of deoxyribonucleotides that is fully complementary to the first string of nucleotides, and

wherein the chimeric RNA-DNA oligonucleotide is [nuclease] RNase protected, and wherein the chimeric RNA-DNA oligonucleotide has [contiguous nucleotides] in each of the first and second strings a set of contiguous nucleotides that are fully complementary to a segment of DNA of the selected gene except that the first and second strings have one, two or four pairs of nucleotide insertions or deletions that defines a site of modification in the selected gene.

19. (Amended) The method of claim 18, wherein the selected gene is tyrosinase, COL7A1, LAMA3, LAMB3, LAMC2, COL17A1, ITGA6, ITGB4, PLEC1, KRT5, KRT14, PKP1, KRT1, KRT10, KRT9, KRT16, LOR, KRT2, KRT6, KRT 16, KRT 17, STS, TGM1, GJB2, GJB3, ATP2A2, DSP, DSG1, HR, hHB1, hHB6, PAX3, TYR, TYRP-1, OCA2, OA1, MITF, HPS, FECH, UROS, URO-D, PPO, XPA, XPB, XPC, XPD, XPG, [CSB,] PTC, STK11/LKB1, PTEN, PTEN, XPB, XPD, WHN, GLA, ATM, ENG, ALK-1, a cytokine BPAG2 or DSG3 gene.

23. (Twice Amended) The method of claim 18, wherein the chimeric RNA-DNA oligonucleotide comprises:

(a) a first string of nucleotides wherein the first string is made of at least four contiguous deoxyribonucleotides flanked on each side by at least nine ribonucleotides; and

(b) a second string of deoxyribonucleotides that is fully complementary to the first string of nucleotides, and

wherein the chimeric RNA-DNA oligonucleotide is [nuclease] RNase protected, and wherein the chimeric RNA-DNA oligonucleotide has [contiguous nucleotides] in each of the first and second strings a set of contiguous nucleotides that are fully complementary to a segment of

DNA of the selected gene except that the first string has one mismatching deoxyribonucleotide in said contiguous deoxyribonucleotides that defines a site of modification in the selected gene.

24. (Twice Amended) The method of claim 18, wherein the chimeric RNA-DNA oligonucleotide comprises:

(a) a first string of nucleotides wherein the first string is made of at least 20 ribonucleotides; and

(b) a second string of deoxyribonucleotides having the same number of deoxyribonucleotides as in the first string of nucleotides, wherein the second string is fully complementary to the first string of nucleotides except that the second string has a deoxyribonucleotide that forms a mismatched base pair with the corresponding nucleotide in the first string to make the genetic modifications in the selected gene, and

wherein the chimeric RNA-DNA oligonucleotide is [nuclease] RNase protected, and wherein the chimeric RNA-DNA oligonucleotide has [contiguous nucleotides] in each of the first and second strings a set of contiguous nucleotides that are fully complementary to a segment of DNA of the selected gene except that the deoxyribonucleotide in the second string also forms a mismatched base pair with the corresponding deoxyribonucleotide in the DNA strand of the selected gene which mismatched base pair defines a site of modification in the selected gene.

25. (Twice Amended) The method of claim 18, wherein the chimeric RNA-DNA oligonucleotide comprises:

(a) a first string of nucleotides wherein the first string is made of at least four contiguous deoxyribonucleotides flanked on each side by at least nine ribonucleotides; and

(b) a second string of deoxynribonucleotides that is fully complementary to the first string of nucleotides, and

wherein the chimeric RNA-DNA oligonucleotide is [nuclease] RNase protected, and wherein the chimeric RNA-DNA oligonucleotide has [contiguous nucleotides] in each of the first and second strings a set of contiguous nucleotides that are fully complementary to a segment of

DNA of the selected gene except that the first and second strings have one, two or four pairs of nucleotide insertions or deletions that defines a site of modification in the selected gene.

33. (Amended) The animal model of claim 32, wherein the selected skin gene is Tyr, COL7A1, LAMA3, LAMB3, LAMC2, COL17A1, ITGA6, ITGB4, PLEC1, KRT5, KRT14, PKP1, KRT1, KRT10, KRT9, KRT16, LOR, 1998, KRT2e, KRT6a, KRT 16, KRT 17, STS, TGM1, GJB2, GJB3, ATP2A2, DSP, DSG1, HR, hHB1, hHB6, PAX3, TYR, TYRP-1, OCA2, OA1, MITF, HPS, FECH, UROS, URO-D, PPO, XPA, XPB, XPC, XPD, XPG, [CSB,] PTC, STK11/LKB1, PTEN, PTEN, XPB, XPD, WHN, GLA, ATM, ENG, ALK-1, a cytokine BPAG2 or DSG3 gene.

APPENDIX: Pending claims in application Serial No. 09/473,872 after the entry of the proposed amendment filed in response to the Office Action dated March 21, 2001:

1. A method of modifying a selected gene in cells of a human skin in vivo which comprises delivering to said cells at one or more locations of the human skin an effective amount of a composition sufficient to bring about stable genetic and phenotypic modifications in the selected gene at said locations wherein the composition comprises a chimeric RNA-DNA oligonucleotide having a double hairpin structure with pyrimidine loops and a pharmaceutically acceptable carrier such that said genetic modifications are made to the selected gene which result in phenotypic changes at said locations of the human skin.

2. The method of claim 1, wherein the stable genetic modification is in an epidermal fragility disorder gene selected from the group consisting of COL7A1, LAMA3, LAMB3, LAMC2, COL17A1, ITGA6, ITGB4, PLEC1, KRT5, KRT14 and PKP1.

3. The method of claim 1, wherein the stable genetic modification is in a keratinization disorder gene selected from the group consisting of KRT1, KRT10, KRT9, KRT16, LOR, KRT2e, KRT6a, KRT 16, KRT 17, STS, TGM1, GJB2, GJB3, ATP2A2, DSP and DSG1.

4. The method of claim 1, wherein the selected gene is tyrosinase, COL7A1, LAMA3, LAMB3, LAMC2, COL17A1, ITGA6, ITGB4, PLEC1, KRT5, KRT14, PKP1, KRT1, KRT10, KRT9, KRT16, LOR, KRT2e, KRT6a, KRT 16, KRT 17, STS, TGM1, GJB2, GJB3, ATP2A2, DSP, DSG1, HR, hHB1, hHB6, PAX3, TYR, TYRP-1, OCA2, OA1, MITF, HPS, FECH, UROS, URO-D, XPA, XPB, XPC, XPD, XPG, PTC, STK11/LKB1, PTEN, PTEN, XPB, XPD, WHN, GLA, ATM, ENG, ALK-1, PPO, BPAG2, or DSG3 gene.

5. The method of claim 1, wherein the selected gene is tyrosinase gene.

6. The method of claim 1, wherein the selected gene is COL7A1 gene.

7. The method of claim 1, wherein the selected gene is KRT17 gene.

8. The method of claim 1, wherein the chimeric RNA-DNA oligonucleotide comprises:

(a) a first string of nucleotides wherein the first string is made of at least four contiguous deoxyribonucleotides flanked on each side by at least nine ribonucleotides; and

(b) a second string of deoxyribonucleotides that is fully complementary to the first string of nucleotides, and

wherein the chimeric RNA-DNA oligonucleotide is, RNase protected, and wherein the chimeric RNA-DNA oligonucleotide has in each of the first and second strings a set of contiguous nucleotides that are fully complementary to a segment of DNA of the selected gene except that the first string has one mismatching deoxyribonucleotide in said contiguous deoxyribonucleotides that defines a site of modification in the selected gene.

9. The method of claim 1, wherein the chimeric RNA-DNA oligonucleotide comprises:

(a) a first string of nucleotides wherein the first string is made of at least 20 ribonucleotides; and

(b) a second string of deoxyribonucleotides having the same number of deoxyribonucleotides as in the first string of nucleotides, wherein the second string is fully complementary to the first string of nucleotides except that the second string has a deoxyribonucleotide that forms a mismatched base pair with the corresponding nucleotide in the first string, and

wherein the chimeric RNA-DNA oligonucleotide is RNase protected, and wherein the chimeric RNA-DNA oligonucleotide has in each of the first and second strings a set of contiguous nucleotides that are fully complementary to a segment of DNA of the selected gene except that the deoxyribonucleotide in the second string also forms a mismatched base pair with

the corresponding deoxyribonucleotide in the DNA strand of the selected gene which mismatched base pair defines a site of modification in the selected gene.

10. The method of claim 1, wherein the chimeric RNA-DNA oligonucleotide comprises:

(a) a first string of nucleotides wherein the first string is made of at least four contiguous deoxyribonucleotides flanked on each side by at least nine ribonucleotides; and

(b) a second string of deoxyribonucleotides that is fully complementary to the first string of nucleotides, and

wherein the chimeric RNA-DNA oligonucleotide is RNase protected, and wherein the chimeric RNA-DNA oligonucleotide has in each of the first and second strings a set of contiguous nucleotides that are fully complementary to a segment of DNA of the selected gene except that the first and second strings have one, two or four pairs of nucleotide insertions or deletions that defines a site of modification in the selected gene.

11. The method of claim 1, wherein the stable genetic modification is correction of a mutation.

12. The method of claim 11, wherein the mutation is a point mutation or a frame shift mutation.

13. The method of claim 1, wherein the stable genetic modification is generation of a mutation.

14. The method of claim 13, wherein the mutation is a point mutation or a frame shift mutation.

15. The method of claim 13, wherein the mutation is a dominant mutation.

16. The method of claim 1, wherein said phenotypic changes include the correction of a skin disorder.

17. The method of claim 1, wherein said phenotypic changes include the correction of albinism, an epidermal fragility disorder or a keratinization disorder.

18. A method of modifying a selected gene in cells of an animal skin in vivo which comprises delivering to said cells at one or more locations of the animal skin an effective amount of a composition comprising a chimeric RNA-DNA oligonucleotide having a double hairpin structure with pyrimidine loops and a pharmaceutically acceptable carrier such that the stable genetic modifications are made to the selected gene which result in phenotypic changes at said locations of the animal skin, wherein the animal is a mouse.

19. The method of claim 18, wherein the selected gene is tyrosinase, COL7A1, LAMA3, LAMB3, LAMC2, COL17A1, ITGA6, ITGB4, PLEC1, KRT5, KRT14, PKP1, KRT1, KRT10, KRT9, KRT16, LOR, KRT2, KRT6, KRT 16, KRT 17, STS, TGM1, GJB2, GJB3, ATP2A2, DSP, DSG1, HR, hHB1, hHB6, PAX3, TYR, TYRP-1, OCA2, OA1, MITF, HPS, FECH, UROS, URO-D, PPO, XPA, XPB, XPC, XPD, XPG, PTC, STK11/LKB1, PTEN, PTEN, XPB, XPD, WHN, GLA, ATM, ENG, ALK-1, a cytokine BPAG2 or DSG3 gene.

20. The method of claim 18, wherein the selected gene is tyrosinase gene.

21. The method of claim 18, wherein the selected gene is COL7A1 gene.

22. The method of claim 18, wherein the selected gene is KRT17 gene.

23. The method of claim 18, wherein the chimeric RNA-DNA oligonucleotide comprises:

(a) a first string of nucleotides wherein the first string is made of at least four contiguous deoxyribonucleotides flanked on each side by at least nine ribonucleotides; and

(b) a second string of deoxyribonucleotides that is fully complementary to the first string of nucleotides, and

wherein the chimeric RNA-DNA oligonucleotide is RNase protected, and wherein the chimeric RNA-DNA oligonucleotide has in each of the first and second strings a set of contiguous nucleotides that are fully complementary to a segment of DNA of the selected gene except that the first string has one mismatching deoxyribonucleotide in said contiguous deoxyribonucleotides that defines a site of modification in the selected gene.

24. The method of claim 18, wherein the chimeric RNA-DNA oligonucleotide comprises:

(a) a first string of nucleotides wherein the first string is made of at least 20 ribonucleotides; and

(b) a second string of deoxyribonucleotides having the same number of deoxyribonucleotides as in the first string of nucleotides, wherein the second string is fully complementary to the first string of nucleotides except that the second string has a deoxyribonucleotide that forms a mismatched base pair with the corresponding nucleotide in the first string to make the genetic modifications in the selected gene, and

wherein the chimeric RNA-DNA oligonucleotide is RNase protected, and wherein the chimeric RNA-DNA oligonucleotide has in each of the first and second strings a set of contiguous nucleotides that are fully complementary to a segment of DNA of the selected gene except that the deoxyribonucleotide in the second string also forms a mismatched base pair with the corresponding deoxyribonucleotide in the DNA strand of the selected gene which mismatched base pair defines a site of modification in the selected gene.

25. The method of claim 18, wherein the chimeric RNA-DNA oligonucleotide comprises:

(a) a first string of nucleotides wherein the first string is made of at least four contiguous deoxyribonucleotides flanked on each side by at least nine ribonucleotides; and

(b) a second string of deoxynribonucleotides that is fully complementary to the first string of nucleotides, and

wherein the chimeric RNA-DNA oligonucleotide is RNase protected, and wherein the chimeric RNA-DNA oligonucleotide has in each of the first and second strings a set of contiguous nucleotides that are fully complementary to a segment of DNA of the selected gene except that the first and second strings have one, two or four pairs of nucleotide insertions or deletions that defines a site of modification in the selected gene.

26. The method of claim 18, wherein the stable genetic modification is correction of a mutation.

27. The method of claim 26, wherein the mutation is a point mutation or a frame shift mutation.

28. The method of claim 18, wherein the stable genetic modification is generation of a mutation.

29. The method of claim 28, wherein the mutation is a point mutation or a frame shift mutation.

30. The method of claim 28, wherein the mutation is a dominant mutation.

31. The method of claim 18, wherein said phenotypic changes include the correction of albinism, an epidermal fragility disorder or a keratinization disorder.

32. An animal model having a skin disorder at one or more locations of its skin wherein the skin disorder is a result of a treatment at said locations with a composition

comprising a chimeric RNA-DNA oligonucleotide having a double hairpin structure with pyrimidine loops targeted to a selected skin gene, said oligonucleotide thereby causing a mutation in the selected skin gene which mutation leads to the skin disorder, wherein the skin disorder is an epidermal fragility disorder, a keratinization disorder or albinism disorder.

33. The animal model of claim 32, wherein the selected skin gene is Tyr, COL7A1, LAMA3, LAMB3, LAMC2, COL17A1, ITGA6, ITGB4, PLEC1, KRT5, KRT14, PKP1, KRT1, KRT10, KRT9, KRT16, LOR, 1998, KRT2e, KRT6a, KRT 16, KRT 17, STS, TGM1, GJB2, GJB3, ATP2A2, DSP, DSG1, HR, hHB1, hHB6, PAX3, TYR, TYRP-1, OCA2, OA1, MITF, HPS, FECH, UROS, URO-D, PPO, XPA, XPB, XPC, XPD, XPG, PTC, STK11/LKB1, PTEN, PTEN, XPB, XPD, WHN, GLA, ATM, ENG, ALK-1, a cytokine BPAG2 or DSG3 gene.

34. The animal model of claim 33, wherein the selected gene is Tyr gene.

35. The animal model of claim 33, wherein the selected gene is COL7A1 gene.

36. The animal model of claim 33, wherein the selected gene is KRT17 gene.

37. The animal model of claim 32, wherein the skin disorder is due to generation of a mutation in the selected skin gene.

38. The animal model of claim 37, wherein the mutation is a point mutation or a frame shift mutation.

39. The animal model of claim 37, wherein the mutation is a dominant mutation.

40. A method of correcting a mutation in a tyrosinase gene in cells of a mammalian skin in vivo which comprises delivering to said cells at one or more locations of the mammalian skin an effective amount of a composition comprising a Tyr-A RNA-DNA oligonucleotide for

causing stable genetic correction in the tyrosinase gene and a pharmaceutically acceptable carrier such that the correction results in restoration of tyrosinase enzyme activity at said locations of the mammalian skin, wherein the mammalian skin is selected from the group consisting of a human and a mouse.